Background: Pancreatic ductal adenocarcinoma (PDAC) features a fibrotic stroma that obstructs immune infiltration. We hypothesize that dysregulated CD26/DPP4 enzymatic activity in PDAC constrains the efficacy of immunotherapy. We also posit that in vivo pharmacological targeting of CD26 enzyme activity, via FDA-approved inhibitors (sitagliptin or STG) for Type 2 Diabetes, influences immune infiltration and enhances immunotherapy response in murine PDAC models.

Methods: CD26 protein was evaluated in immortalized cancer associated fibroblasts (CAFs) and PDAC cells by immunoblot and flow, while soluble CD26 was measured by ELISA. In vivo studies used C57BL/6 mice orthotopically implanted with syngeneic luciferase-expressing KPC-tumor cells in the pancreas. Tumors were verified and monitored by bioluminescence imaging (BLI) and mice randomized to treatment: STG (75mg/kg in drinking water), anti-PD-L1 Ab (200ug 3x/week), or both for 9 days. Controls received vehicle and/or isotype Ab. Tumors were harvested at endpoint and tumor infiltrating lymphocytes (TIL) and checkpoint expression was evaluated. Two in vivo studies evaluated survival (treated 3 weeks) or TIL (treated 9 days) for PDAC-bearing mice treated with STG, anti-PD-L1, and anti-Lag3 (including single, double, and triple therapy combinations). A combination study evaluating survival in mice orthotopically implanted with MT5 cells is ongoing.

Results: CD26 was among the most highly expressed genes in patient PDAC-derived CAFs vs. normal fibroblasts. CD26 protein was observed in human CAFs (HT137 and h-iPSC-PDAC-1) and PDAC cells (HPAC and PANC1) via immunoblot and flow, however, soluble CD26 was only observed in CAF supernatants (180 pg/mL and 587.6 pg/mL). Concurrent administration of STG and anti-PD-L1 Ab limited tumor progression and increased CD4, as well as increased CD8 (p's<0.04). CD4+ TIL from STG+anti-PD-L1 had high Lag-3 expression (p's<0.025). Triple therapy (STG+anti-PD-L1 Ab, and anti-Lag-3) increased overall survival (p=0.005) of mice bearing KPC-luc tumors and increased CD3 vs all groups (p's<p=0.02). Our ongoing survival study in mice bearing MT5 tumors initially demonstrates similar benefit from treatment with STG administered in combination with immune checkpoint blockade.

Conclusions: Our results are the first to demonstrate CD26 enzyme inhibition (STG), augments in vivo efficacy of anti-PD-L1 Ab and anti-Lag-3 in relevant orthotopic PDAC models. This work identifies immune populations, including T cells, that provide mechanistic insight for efficacy. We demonstrate versatility of CD26 inhibition and its capacity to modulate checkpoint molecules and efficacy in PDAC.

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